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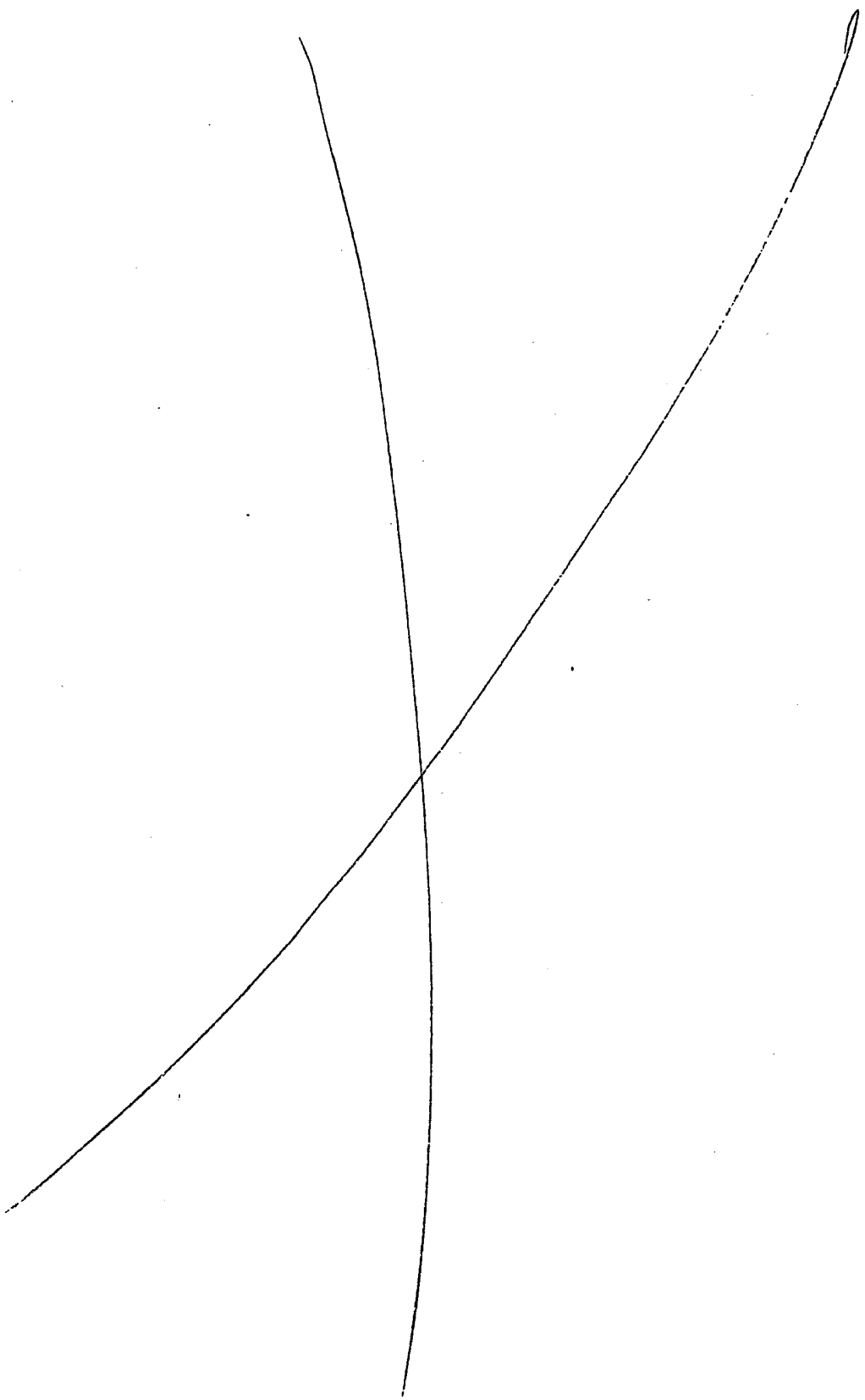

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Carbon monoxide source for the preparation of transition metal carbonyl complexes

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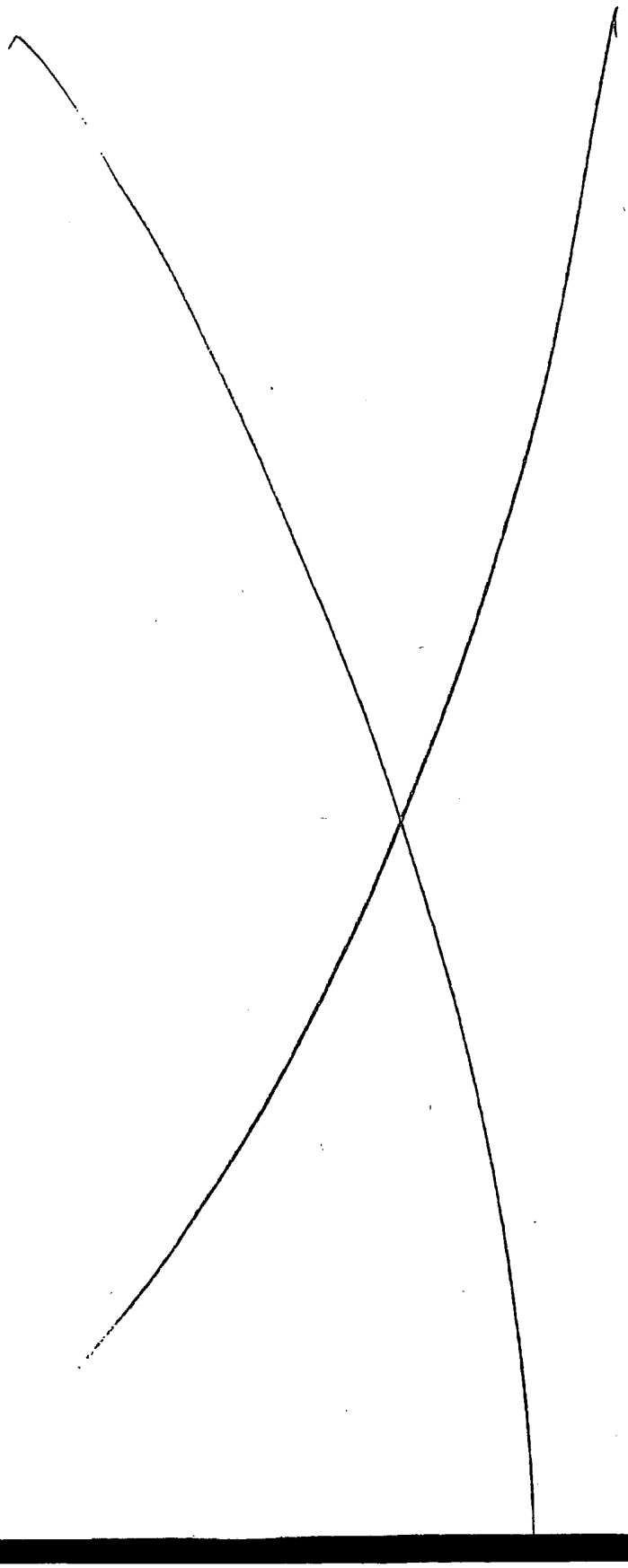

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CARBON MONOXIDE SOURCE FOR THE PREPARATION OF TRANSITION METAL CARBONYL COMPLEXES

The present invention relates to compounds that have a novel use as a carbon monoxide source and optionally as a reducing agent in the preparation of transition metal carbonyl complexes.

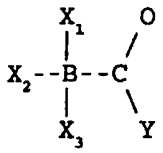
Carbonyl complexes are compounds that contain carbon monoxide as a coordinated ligand. Carbon monoxide is a common ligand in transition metal chemistry, in part due to the synergistic nature of its bonding to transition metals.

The bonding of CO to a metal consists of two components. The first component of the bonding is based on σ-donation, the overlap of a lone pair on the carbon atom with an empty d-orbital of the metal. The second component consists in π-back-donation from a filled d-orbital of the metal into an empty π^* orbital of the carbon atom of CO. This second component is called pi-backbonding or pi-backdonation.

The above described formation of carbonyl complexes with transition metals is crucial for the application of such compounds in the labeling of proteins, peptides and a variety of other compounds. For many applications these molecules are labeled by means of a so-called labeling kit which contains the necessary reagents. Current kits are based on boronhydride as the reducing agent, further contain tartrate, lactose and borate buffer, pH 11.5, and are filled with gaseous CO as the CO source. The disadvantages of these known reaction mixtures are the slow dissolution of CO into the reaction solvent resulting in a decreased yield of carbonyl complexes, the impossibility of industrial preparation of large amounts of CO filled kit vials and the slow diffusion of CO even through tightly closed vials. Moreover, the pH is rather high, which is not convenient.

It is the object of the present invention to provide an alternative for CO and sodium boron hydride that does not have the above stated drawbacks.

It has now been found that compounds of formula I



(I)

wherein:

X_1 is -H;

X_3 and X_2 are substituents which may be the same or different and are selected from the group consisting of -H, $-NH_xR_y$ with x+y=3, or -R, wherein R is a substituent which is bound by a carbon atom to the nitrogen or boron, respectively, and is preferably alkyl or aryl;

Y is -OH, $-OH_2$, -OR or -NHR, wherein R is a substituent which is bound by a carbon atom to the nitrogen or oxygen, respectively, and is preferably alkyl or aryl;

or salts thereof

can be used as a carbon monoxide (CO) source and optionally also as a reducing agent in the preparation of metal carbonyl complexes in aqueous solution. If Y is -OH or $-OH_2$, the compounds are acids which can be deprotonated (i.e. with NaOH). In that case, the compounds which are isolated are the salts (boroncarbonate anion $R_3B\text{-}COO^{2-}$ plus the corresponding cation, e.g. Li^+, Na^+, Ca^{2+}, Mg^{2+} and others). The reducing agent function is only present if at least one of X_1, X_2 and X_3 is a hydrogen. For stability reasons it is preferred that two of X_1, X_2 and X_3 are -H. The carbon monoxide is released upon heating an aqueous solution of the compound.

The advantages of the above compounds are the following. CO is produced for the first time in aqueous media under controllable conditions (pH, temperature). Carbonyl complexes of the claimed metals can be prepared in water at well defined conditions instead of organic

solvents or under high pressure and high temperature. The CO source and reducing agent can be present in the same single compound, which is convenient since reduction is practically always required for the preparation of carbonyls. In case the metal to be complexed is Tc-99m or Re-188/186 kits can be produced without the demand of filling a vial with toxic and volatile CO. A major advantageous embodiment is a molecule combining the different functionalities in one compound. Such compound can act as a reducing agent and as an in situ CO source, where the CO is only produced if a protic solvent (like water) is present.

By varying the substituents at the different positions various types of compounds can be obtained. These can be subdivided in the following groups:

1. a borane carbonate compound in which X_1, X_2 and X_3 are -H and Y is $-OH_2$, and/or the corresponding salts of the mono- or dideprotonated borane carbonate $[H_3BCO_2]^{2-}$;
2. a borane amino acid (ammonia carboxy borane) in which X_1 is NH_3, X_2 and X_3 are -H and Y is -OH, and/or the corresponding salts of the monodeprotonated ammine borane carbonate $[(NH_3)H_2BCO_2]^-$;;
3. alkylated borane amino acids (trialkyl ammonia carboxy boranes) in which X_1 is $-NH_xR_y$ with x+y=3, wherein R is a substituent which is bound by a carbon atom to the nitrogen and is preferably alkyl or aryl, X_2 and X_3 are -H and Y is -OH.
4. compounds of formula I wherein X_1 is an organic substituent bound by a carbon atom to boron, X_2 and X_3 are -H and Y is $-OH_2$.
5. compounds of formula I wherein X_1 and X_2 are organic substituents bound by a carbon atom to boron, X_3 is -H and Y is $-OH_2$.
6. borane carboxylic acid alkylester compounds wherein X_1, X_2 and X_3 are as defined under 1-5 above and Y is OR', in which R' is a substituent bound by a carbon atom to the oxygen, such as an alkyl, more in particular methyl or ethyl.

7. borane carbamate compounds wherein X_1, X_2 and X_3 are as defined in 1-5 above and Y is NH_2, NHR" or $NR"_2$, wherein R" is a substituent bound by a carbon atom to nitrogen, such as an alkyl, more in particular methyl or ethyl.

Particular examples of these compounds are:
boranocarbonate derivatives: $[H_3B\text{-}COOH_2]$, $[H_3B\text{-}COOH]M$, $[H_3B\text{-}COO]M_2$, $Na[H_3B\text{-}COOCH_3]$, wherein M is an alkali cation;
boranocarbamates: $Na[H_3BCONHCH_3]$, $M[H_3B\text{-}CONH_2]$, wherein M is an alkali cation;
ammine-boranocarbonates: $[H_3N\text{-}BH_2\text{-}COOH]$, $[H_3N\text{-}BH_2\text{-}COO]Li$, $[(CH_3)_3N\text{-}BH_2\text{-}COOH]$, $[(CH_3)H_2N\text{-}BH_2\text{-}COOH]$, $[(CH_3)H_2N\text{-}BH_2\text{-}COO]Li$, $[(CH_3)H_2N\text{-}BH_2\text{-}COOCH_3]$;
ammine-boranocarbamates: $[H_3N\text{-}BH_2\text{-}CONH_2]$, $[(CH_3)_2HN\text{-}BH_2\text{-}CONHC_2H_5]$

The compounds of the invention can be prepared by means of or analogous to the methods as described by Burg et al., J. Am. Chem. Soc. 59, 780 (1936) for BH_3CO; Malone et al., Inorg. Chem. 6, 817 (1967) for $M_2[H_3B\text{-}COO]$ and $M[H_3B\text{-}COOC_2H_5]$; Howe et al., Inorg. Chem. 10, 930 (1971) for $M[H_3B\text{-}CONH_2]$; Spielvogel et al., J. Am. Chem. Soc. 102, 6343 (1980) for $[H_3N\text{-}BH_2\text{-}COOH]$ and $[(CH_3)_3N\text{-}BH_2\text{-}CONHC_2H_5]$; Spielvogel et al., Inorg. Chem. 23, 4322 (1984) for $[(CH_3)H_2N\text{-}BH_2\text{-}COOCH_3]$; Spielvogel et al., Inorg. Chem. 23, 1776 (1984) and J. Am. Chem. Soc. 98, 5702 (1976) for $[H_3N\text{-}BH_2\text{-}CONH_2]$, $[(CH_3)_2HN\text{-}BH_2\text{-}CONHC_2H_5]$.

The invention further relates to a method for preparing transition metal carbonyl complexes, wherein one or more of the compounds defined above are used as the CO source and optionally as the reducing agent. This method comprises in summary the release of CO from any compound of the invention, in particular from one or more of the compounds 1-7, in water or buffer due to hydrolytic reactions. Concomitantly, the metal with which a carbonyl should be formed is reduced by the hydride substituent attached to boron. The compounds of the invention, in particular compounds 1-7, are dissolved in water or buffer and the metal is added either as a solid

or as a solution. Hydrolysis of the compounds of the invention, in particular of compounds 1-7, releases CO. At the same time, the hydrides attached to the boron (-H) will reduce the metal center to a valency where the metal is able to coordinate the released CO. In that moment, carbonyl complexes are formed. The method according to the invention for preparing carbonyl complexes, thus comprises mixing the boron compounds of the invention with an aqueous solution of the metal.

The compounds and method of the invention are suitable for the formation of any carbonyl complex, but in particular those in which the transition metal in the transition metal carbonyl complex is selected from the groups V-B to VIII-B metals. More in particular the method is suitable for preparing carbonyl complexes of the following transition metals: Vanadium (V), Chromium (Cr), Molybdenum (Mo), Tungsten (W), Manganese (Mn), Technetium (Tc), Rhenium (Re), Iron (Fe), Ruthenium (Ru), Osmium (Os), Cobalt (Co), Rhodium (Rh), Iridium (Ir) and Nickel (Ni).

Furthermore, the invention provides a kit for preparing transition metal carbonyl complexes, comprising a compound according to the invention in aqueous solution, a stabilizing agent like tartrate, glucoheptonate, lactate, citrate and a buffer system like borate or phosphate. In a preferred embodiment thereof, the kit of the invention contains at least 2 mg borane carbonate, preferably in a borate buffer (pH 9.1) in an oxygen-free environment under a nitrogen atmosphere. It is preferred that the total volume of the solution after addition of the radioactive metal solution does not exceed 1 ml. Suitable incubation conditions comprise heating the solution for about 20 minutes to 75°C.

The use of the compounds according to the invention is more broadly applicable than solely for the preparation of carbonyl complexes, but can also be applied in other circumstances wherein a CO source in aqueous solution is required.

The present invention is further illustrated in the following examples, that are given for illustration purposes only.

EXAMPLES

EXAMPLE 1

Preparation of $K_2H_3BCO_2$

1. Synthesis of $BH_3 \cdot CO$

4 g of $NaBH_4$ was carefully added to 15 ml of concentrated H_3PO_4 (dried overnight under high vacuum at room temperature) in vacuo (1 mbar) under vigorous stirring over a period of 2 hours. The evolving BH_3 was dried by passing it through a cool trap at -78°C and was condensed in a second cool trap at -200°C containing 70 ml of dry DME. The second trap was disconnected from the first trap and the vacuum line. The temperature was brought to -40°C. Subsequently the trap was pressurized with 1.3 bar of dry CO. The reaction mixture was stirred in a cool bath at -40°C (dry ice with acetonitrile) under 1.3 bar of CO overnight.

2. Synthesis of $K_2H_3BCO_2$

The gas outlet of the trap was connected to a 100 ml two-neck round-bottom flask (equipped with gas inlet and reflux condenser) containing 50 ml of dry ethanol and 3 g KOH. The cool bath of the trap was removed and the evolving $BH_3 \cdot CO$ was bubbled slowly through the ethanolic KOH solution at 0°C. The DME solution was slowly heated to 80°C and the trap subsequently three times flushed with CO. After the evolution of $BH_3 \cdot CO$ had stopped the ethanolic solution was refluxed for 30 min. After cooling the solution to room temperature $K_2H_3BCO_2$ precipitated as a white powder which was filtered by a sintered glass filter, washed with ice cold ethanol and dried under vacuum.

EXAMPLE 2

Labeling experiment using a lyophilized kit

A labeling kit was prepared by lyophilizing 1 mg $K_2[BH_3COO]$ in 0.1 ml of 0.1M PBS, pH 7.5 in a vial that was flushed with N_2. As an alternative a 0.1M borate buffer, pH 8.5 can be used.

For labeling, 1 ml of a generator eluted $[^{99m}TcO_4]^-$ saline solution is added. It was found that the yield is independent of the absolute amount of $[^{99m}TcO_4]^-$. The solution thus obtained is heated to 75°C for 20 min.

The yields are between 80 and 100% (trace 1 in Fig. 1) for pH 7.5; trace 3 in Fig. 1 for pH 8.5.

To establish the identity of the compound, picolinic acid was added directly to the reaction solution, in which the carbonyl complex had been prepared. HPLC revealed the complex $[^{99m}Tc(OH_2)(pic)(CO)_3]$ (Fig. 1, trace 2) by comparison with "cold", i.e. non-labeled material. The "hot" material is found by means of a radioactivity detector, whereas the "cold" material is detected with a UV detector.

EXAMPLE 3

Labeling experiment with a so-called "wet kit"

A vial containing 2 mg borane carbonate and a generator eluate of pertechnetate in borate buffer, pH 9.1, in a total volume of 1 ml was heated for 20 min. to 75°C. The labeling yield thus obtained was higher than 97%.